

Dean L. Engelhardt, et al.

Serial No.: 08/486,069

Filed: June 7, 1995

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Third Supplemental Amendment, Their February 2, 1999 Second Supplemental

Amendment, Their July 24, 1998 Supplemental Response and Their July 6, 1998

Amendment Under 37 C.F.R. §1.116 - May 1, 1999)



**AMEND THE ABOVE-IDENTIFIED APPLICATION AS FOLLOWS:**

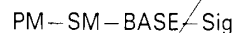
**In The Claims:**

Amend claims 284, 314, 315, 329, 331, 332, 337, 348, 373, 376, 377, 381, 385 and 390 as follows:

284. (Five Times Amended) A process for detecting a nucleic acid of interest in a sample, which process comprises the steps of:

(a) hybridizing said nucleic acid of interest in the sample with [an] one or more oligo- or [polynucleotide] polynucleotides, each such oligo- or polynucleotide being complementary to or capable of hybridizing with said nucleic acid of interest or a portion thereof, said oligo- or polynucleotide comprising at least one nucleotide selected from the group consisting of:

(i) a nucleotide having the formula



wherein

PM is a phosphate moiety,

SM is a [monosaccharide] furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is a detectable moiety,

wherein PM is attached to the furanose moiety SM at a position selected from the group consisting of the 2', the 3' [or] and the 5' position, or any combination thereof [of the monosaccharide moiety SM when said nucleotide is a deoxyribonucleotide and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide], BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is a purine or a 7-deazapurine, and Sig is covalently attached to BASE at a position other

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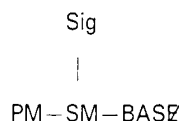
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than the C<sup>5</sup> position when BASE is a pyrimidine, at a position other than the C<sup>8</sup> position when BASE is a purine and at a position other than the C<sup>7</sup> position when BASE is a 7-deazapurine and such covalent attachment does not substantially interfere with double helix formation or nucleic acid hybridization;

(ii) a nucleotide having the formula



wherein

PM is a phosphate moiety,

SM is a [monosaccharide] furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is a detectable moiety,

[wherein PM is a phosphate moiety, SM is a monosaccharide moiety, and BASE is a pyrimidine, purine or 7-deazapurine moiety,] said PM being attached to the furanose moiety SM at a position independently selected from the group consisting of the 2', 3', and 5' positions, or any combination thereof [of SM when said nucleotide is a ribonucleotide, and at a position independently selected from the 3' and 5' positions when said nucleotide is a deoxyribonucleotide], said BASE being attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is a purine or 7-deazapurine, and Sig is covalently attached to SM directly or through a linkage group and such covalent attachment does not substantially interfere with double helix formation or nucleic acid hybridization; and

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(iii) a nucleotide having the formula

Sig—PM—SM—BASE

wherein

PM is a phosphate moiety,

SM is a [monosaccharide] furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is a detectable moiety

wherein PM is attached to the furanose moiety SM at a position selected from the group consisting of the 2', the 3' [or] and the 5' position, or any combination thereof [of SM when said nucleotide is a deoxyribonucleotide and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide], BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is a purine, and Sig is covalently attached to PM and such covalent attachment does not substantially interfere with double helix formation or nucleic acid hybridization; and

(b) detecting the presence of said detectable Sig moieties in any of the oligo- or polynucleotides which have hybridized to said nucleic acid of interest.

Claim 314, line 1, after "according to" and before "wherein" change "claim 313" to -- claims 312 or 313 -- .

Claim 315, line 1, after "according to" and before "wherein" change "claim 314" to -- claims 312 or 313 -- .

329. (Twice Amended) A process for determining the sequence of a nucleic acid of interest, comprising the steps of:

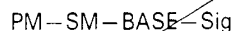
M2 [incorporating one or more modified nucleotides or an oligo- or polynucleotide comprising one or more modified nucleotides into a nucleic acid or] providing labeled nucleic acid fragments, each fragment being complementary to a portion of or to said nucleic acid of interest, wherein each of said fragments comprise one or more modified nucleotides [comprise a] said modified nucleotide or nucleotides being modified on the sugar, phosphate or base moieties thereof, and [wherein said one or more modified nucleotides are] comprising detectable or self-indicating labels [, to produce a labeled nucleic acid or labeled nucleic acid fragments complementary to said nucleic acid of interest or a portion thereof];

[separating said labeled nucleic acid or labeled nucleic acid] subjecting said labeled fragments [in] to a sequencing gel to separate or resolve said fragments; and

detecting the presence of each of said separated or resolved [labeled nucleic acid or labeled nucleic acid fragment] fragments by means of said detectable or self-indicating [modified nucleotide or nucleotides] labels, thereby determining the sequence of said nucleic acid of interest.

Sub 329 331. (Thrice Amended) The process according to claims 329 or 373, wherein said modified nucleotide or nucleotides [comprises] comprise a member selected from the group consisting of:

(ii) a nucleotide having the formula



wherein

PM is a phosphate moiety,

SM is a [monosaccharide] furanose moiety,

BASE is a pyrimidine, purine 7-deazapurine, and

Sig is a detectable moiety,

wherein PM is attached to the furanose moiety SM at a position selected from the group consisting of the 2', [at] the 3' [or] and the 5' position, or

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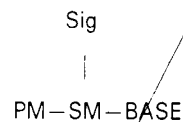
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any combination thereof [of the monosaccharide moiety SM when said nucleotide is a deoxyribonucleotide and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide], BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is a purine or a 7-deazapurine, and Sig is covalently attached to BASE at a position other than the C<sup>5</sup> position when BASE is a pyrimidine, at a position other than the C<sup>8</sup> position when BASE is a purine, and at a position other than the C<sup>7</sup> position when BASE is a 7-deazapurine;

(ii) a nucleotide having the formula



wherein

PM is a phosphate moiety,

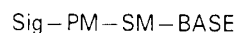
SM is a [monosaccharide] furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is a detectable moiety,

said PM being attached to the furanose moiety SM at a position [independently] selected from the group consisting of the 2', 3', and 5' positions, or any combination thereof [of SM when said nucleotide is a ribonucleotide, and at a position independently selected from the 3' and 5' positions when said nucleotide is a deoxyribonucleotide], said BASE being attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is a purine or 7-deazapurine, and Sig is covalently attached to SM directly or through a linkage group; and

(iii) a nucleotide having the formula



wherein

PM is a phosphate moiety,

SM is a [monosaccharide] furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

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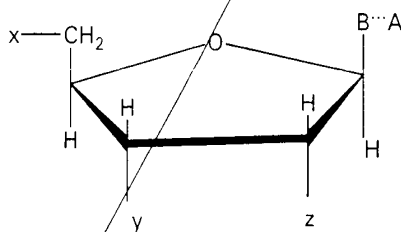
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Sig is detectable moiety,

wherein PM is attached to the furanose moiety SM at a position selected from the group consisting of the 2', the 3' [or] and the 5' position or any combination thereof [of SM when said nucleotide is a deoxyribonucleotide and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide], BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is a purine, and Sig is covalently attached to PM.

332. (Thrice Amended) The process according to claims 329 or 373, wherein said modified nucleotide or nucleotides [has] have the structure:



wherein B represents a purine, a 7-deazapurine or a pyrimidine moiety suitable for incorporation into a polynucleotide and covalently bonded to the C<sup>1</sup>-position of the [monosaccharide] furanose moiety, provided that when B is a purine or 7-deazapurine, the [monosaccharide] furanose moiety is attached at the N<sup>9</sup> position of the purine or deazapurine, and when B is a pyrimidine, the [monosaccharide] furanose moiety is attached at the N<sup>1</sup> position of the pyrimidine;

wherein A represents at least three carbon atoms and is an indicator molecule that is self-indicating;

wherein B and A are covalently attached directly or through a linkage group, said linkage group not interfering substantially with detection of A;

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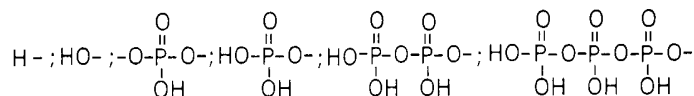
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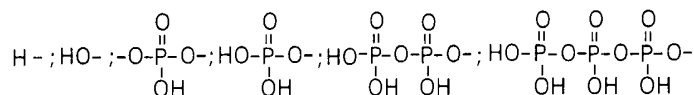
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wherein if B is a purine, A is attached to the 8-position of the purine, if B is a 7-deazapurine, A is attached to the 7-position of the deazapurine, and if B is a pyrimidine, A is attached to the 5-position of the pyrimidine; and

wherein x comprises a member selected from the group consisting of:



wherein y comprises a member selected from the group consisting of:



wherein z comprises a member selected from the group consisting of H- and HO-.

337. (Four Times Amended) A process for preparing a labeled oligo- or polynucleotide of interest, comprising the steps of:

(A) providing either:

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(1) one or more chemically modified nucleotides capable of incorporating into an oligo- or polynucleotide of interest, alone or in conjunction with one or more other modified or unmodified nucleic acids selected from the group consisting of nucleotides, oligonucleotides and polynucleotides, said other modified or unmodified nucleic acids being capable of incorporating into an oligo- or polynucleotide of interest, said chemical modification comprising a label capable of providing directly or indirectly a detectable signal indicating the presence of said labeled oligo- or polynucleotide of interest, or

(2) an oligo- or polynucleotide of interest comprising one or more chemically modified nucleotides, alone or in conjunction with one or more other modified or unmodified nucleic acids selected from the group consisting of nucleotides, oligonucleotides and polynucleotides, said chemical modification comprising a label capable of providing directly or indirectly a

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detectable signal indicating the presence of said labeled oligo- or polynucleotide of interest.

said chemically modified nucleotides being modified on the sugar, phosphate or base moieties thereof and being selected from the group consisting of:

(i)

PM-SM-BASE-Sig

wherein

PM is a phosphate moiety,

SM is a [monosaccharide] furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is a detectable moiety, and

wherein PM is attached to the furanose moiety SM at a position selected from the group consisting of the 2', [at] the 3' [or] and the 5' position, or any combination thereof [of the monosaccharide moiety SM when said nucleotide is a deoxyribonucleotide and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide], BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is a purine or a 7-deazapurine, and Sig is covalently attached to BASE directly or through a linkage group at a position other than the C<sup>5</sup> position when BASE is a pyrimidine, at a position other than the C<sup>8</sup> position when BASE is a purine, and at a position other than the C<sup>7</sup> position when BASE is a 7-deazapurine;

(ii)

Sig

|

PM-SM-BASE

wherein

PM is a phosphate moiety,

SM is a [monosaccharide] furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and



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Sig is a detectable moiety, and wherein said PM is attached to the furanose moiety SM at a position [independently] selected from the group consisting of the 2', 3', and 5' positions, or any combination thereof [of SM when said nucleotide is a ribonucleotide, and at a position independently selected from the 3' and 5' positions when said nucleotide is a deoxyribonucleotide], said BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is a purine or 7-deazapurine, and Sig is covalently attached to SM directly or through a linkage group; and

(iii)

Sig-PM-SM-BASE

wherein

PM is a phosphate moiety,

SM is a [monosaccharide] furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is detectable moiety; and

wherein PM is attached to the furanose moiety SM at a position selected from the group consisting of the 2', the 3' [or] and the 5' position, or any combination thereof [of SM when said nucleotide is a deoxyribonucleotide and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide], BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is purine, and Sig is covalently attached to PM directly or through a linkage group; and

said oligo- or polynucleotide of interest; and

(B) either incorporating said one or more modified nucleotides (1) into said oligo- or polynucleotide, thereby preparing a labeled oligo- or polynucleotide of

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interest, or preparing said oligo- or polynucleotide of interest from said oligo- or polynucleotide (2).

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348. A process for [detecting] determining in a sequencing gel the presence of [an oligo- or polynucleotide] nucleic acid fragments complementary to a nucleic acid of interest or a portion thereof [in a sequencing gel], said process comprising the steps of:

(A) providing:

[(a)] one or more chemically modified nucleotides capable of incorporating into a nucleic acid [an oligo- or polynucleotide], alone or in conjunction with one or more other modified or unmodified nucleic acids selected from the group consisting of nucleotides, oligonucleotides and polynucleotides, said other modified or unmodified nucleic acids being capable of incorporating into or forming one or more nucleic acid fragments, each fragment being complementary to said nucleic acid of interest or to a portion thereof [an oligo- or polynucleotide], said chemical modification rendering said one or more chemically modified nucleotides either:

(I) self-indicating; or

(II) comprising a label capable of providing directly or indirectly a detectable signal;

said self-indicating chemical modification or said label indicating the presence of said labeled [oligo- or polynucleotide] nucleic acids or nucleic acid fragments;

[thereby indicating the presence of said labeled oligo- or polynucleotide,] said chemically modified nucleotides being modified non-disruptively or disruptively on at least one of the sugar, phosphate or base moieties thereof; and

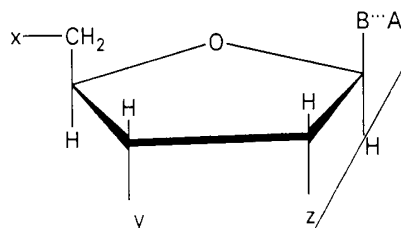
[(b)] an oligo- or polynucleotide];

(B) incorporating said one or more chemically modified nucleotides into said [oligo- or polynucleotide] one or more fragments, thereby preparing [a] labeled [oligo- or polynucleotide] fragments, each such fragment being complementary to

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said nucleic acid of interest or to a portion thereof, said labeled [oligo- or polynucleotide of interest] fragments comprising one or more chemically modified nucleotides selected from the group consisting of:

(i)

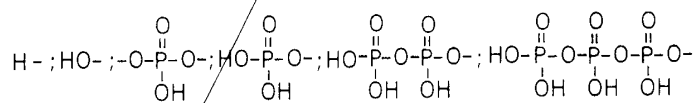


wherein B represents a purine, a 7-deazapurine or a pyrimidine moiety covalently bonded to the C1'-position of the sugar moiety, provided that whenever B is a purine or 7-deazapurine, the sugar moiety is attached at the N9-position of the purine or 7-deazapurine, and whenever B is a pyrimidine, the sugar moiety is attached at the N1-position of the pyrimidine;

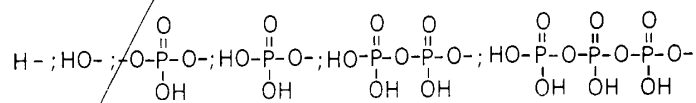
wherein A comprises at least three carbon atoms and represents at least one component of a signalling moiety capable of producing directly or indirectly a detectable signal or being self-indicating; and

wherein B and A are covalently attached directly or through a linkage group, and

wherein x comprises a member selected from the group consisting of:



wherein y comprises a member selected from the group consisting of:



wherein z comprises a member selected from the group consisting of H- and HO-;

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(ii)

Sig

I

PM SM BASE

wherein

PM is a phosphate moiety,

SM is a [monosaccharide] furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is a detectable moiety or is self-indicating, and

wherein said PM is attached to the furanose moiety SM at a position [independently] selected from the group consisting of the 2', 3', and 5' positions, or any combination thereof [of SM when said nucleotide is a ribonucleotide, and at a position independently selected from the 3' and 5' positions when said nucleotide is a deoxyribonucleotide], said BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is a purine or 7-deazapurine, and Sig is covalently attached to SM directly or through a linkage group; and

(iii)

Sig-PM-SM-BASE

wherein

PM is a phosphate moiety,

SM is a [monosaccharide] furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is detectable moiety or is self-indicating; and

wherein PM is attached to the furanose moiety SM at a position selected from the group consisting of the 2', the 3' [or] and the 5' position, or any combination thereof [of SM when said nucleotide is a deoxyribonucleotide and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide], BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is purine, and Sig is covalently attached to PM directly or through a linkage group;

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(C) transferring or subjecting said labeled [oligo- or polynucleotide of interest] fragments to a sequencing gel;

(D) separating or resolving said labeled fragments [oligo- or polynucleotide of interest from other nucleic acids not of interest]; and

(E) detecting directly or indirectly the presence of said labeled [oligo- or polynucleotide] fragments.

373. (Amended) A process for determining the sequence of a nucleic acid of interest, comprising the steps of:

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providing or generating [a labeled nucleic acid or] labeled nucleic acid fragments complementary to said nucleic acid of interest or to a portion thereof, each of said [labeled nucleic acid or labeled nucleic acid] labeled fragments [being] comprising one or more modified nucleotides which comprise detectable or self-indicating labels and [comprising] said one or more modified nucleotides being modified on the sugar, phosphate or base moieties thereof;

introducing or subjecting said [labeled nucleic acid or labeled nucleic acid] fragments [into] to a sequencing gel;

separating or resolving said [labeled nucleic acid or labeled nucleic acid] fragments in said sequencing gel; and

detecting each of the separated or resolved [labeled nucleic acid or labeled nucleic acid] fragments; thereby determining the [polynucleotide] sequence of said nucleic acid of interest [from the labeled nucleic acid or labeled nucleic acid fragments detected].

376. (Amended) A process for determining whether the number of copies of a particular chromosome in a cell is normal or abnormal, the process comprising the steps of:

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contacting said cell under hybridizing conditions with [an] one or more oligo- or [polynucleotide] polynucleotides, each of which is capable of hybridizing

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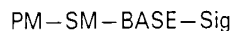
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specifically to a locus or loci of said particular chromosome or a portion thereof, said oligo- or polynucleotide comprising at least one modified nucleotide selected from the group consisting of:

(i) a nucleotide having the formula



wherein

PM is a phosphate moiety,

SM is a furanose moiety,

BASE is a pyrimidine, purine 7-deazapurine, and

Sig is a detectable moiety,

wherein PM is attached to the furanose moiety SM at a position selected from the group consisting of the 2', [at] the 3' [or] and the 5' position, or any combination thereof [of SM when said nucleotide is a deoxyribonucleotide and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide], BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is a purine or a 7-deazapurine, and Sig is covalently attached to BASE at a position other than the C<sup>5</sup> position when BASE is a pyrimidine, at a position other than the C<sup>8</sup> position when BASE is a purine, and at a position other than the C<sup>7</sup> position when BASE is a 7-deazapurine;

(ii) a nucleotide having the formula

Sig



wherein

PM is a phosphate moiety,

SM is a furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is a detectable moiety,

wherein PM is attached to the furanose moiety SM at a position [independently] selected from the group consisting of the 2', 3', and 5' positions, or any combination thereof [of SM when said nucleotide is a

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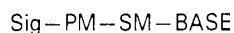
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ribonucleotide, and at a position independently selected from the 3' and 5' positions when said nucleotide is a deoxyribonucleotide], BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is a purine or 7-deazapurine, and Sig is covalently attached SM directly or through a linkage group; and

(iii) a nucleotide having the formula



wherein

PM is a phosphate moiety,

SM is a furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is detectable moiety,

wherein PM is attached to the furanose moiety SM at position selected from the group consisting of the 2', the 3' [or] and the 5' position, or any combination thereof [of SM when said nucleotide is a deoxyribonucleotide

and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide],

BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is purine, and Sig is covalently attached to PM,

to permit hybridization of said oligo- or polynucleotide to the locus or loci of said particular chromosome;

detecting the signal generated by said hybridized oligo- or polynucleotide, thereby determining the number of copies of said particular chromosome; and

comparing said determined number of copies of said particular chromosome with a number of copies of said particular chromosome determined for a normal cell containing said particular chromosome, thereby determining whether the number of copies of said particular chromosome in said cell is abnormal.

377. (Amended) The process of claim 376, wherein said one or more oligo- or [polynucleotide] polynucleotides [comprises] comprise a clone or clones or DNA fragments derived from said particular chromosome.

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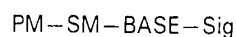
Filed: June 7, 1995

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381. (Amended) A process for identifying a chromosome of interest in a cell containing other chromosomes, the process comprising the steps of:

providing a set of [clones] oligo- or polynucleotides, each of which is specifically [complementary to at least one sequence] hybridizable to a locus or loci in said chromosome of interest, each of said [clones] oligo- or polynucleotides comprising at least one modified nucleotide selected from the group consisting of:

- (i) a nucleotide having the formula



wherein

PM is a phosphate moiety,

SM is a furanose moiety,

BASE is a pyrimidine, purine 7-deazapurine, and

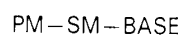
Sig is a detectable moiety,

wherein PM is attached to the furanose moiety SM at a position selected from the group consisting of the 2', [at] the 3' [or] and the 5' position, or any combination thereof [of SM when said nucleotide is a deoxyribonucleotide and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide], BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>3</sup> position when BASE is a purine or a 7-deazapurine, and Sig is covalently attached to BASE at a position other than the C<sup>5</sup> position when BASE is a pyrimidine, at a position other than the C<sup>8</sup> position when BASE is a purine, and at a position other than the C<sup>7</sup> position when BASE is a 7-deazapurine;

- (ii) a nucleotide having the formula

Sig

|



wherein

PM is a phosphate moiety,

SM is a furanose moiety,



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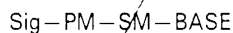
Sub  
to  
be  
AM  
cont

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is a detectable moiety,

wherein PM is attached to the furanose moiety SM at a position  
[independently] selected from the group consisting of the 2', 3' and 5'  
positions, or any combination thereof [of SM when said nucleotide is a  
ribonucleotide, and at a position independently selected from the 3' and 5'  
positions when said nucleotide is a deoxyribonucleotide], BASE is attached  
to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or  
the N<sup>9</sup> position when BASE is a purine or 7-deazapurine, and Sig is covalently  
attached SM directly or through a linkage group; and

(iii) a nucleotide having the formula



wherein

PM is a phosphate moiety,

SM is a furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is detectable moiety,

wherein PM is attached to the furanose moiety SM at a position selected  
from the group consisting of the 2', the 3' [or] and the 5' position, or any  
combination thereof [of SM when said nucleotide is a deoxyribonucleotide  
and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide],  
BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is  
a pyrimidine or the N<sup>9</sup> position when BASE is purine, and Sig is covalently  
attached to PM;

fixing the chromosomes from or in said cell;

contacting said fixed chromosomes under hybridizing conditions with said

[clones] oligo- or polynucleotides, permitting hybridization of said [clones] oligo- or  
polynucleotides to said [complementary sequence] locus or loci in said chromosome  
of interest;

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Sub  
A8  
Q8  
cont

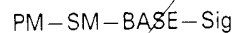
detecting any signal generated by each of said ~~[clones]~~ oligo- or polynucleotides which have hybridized to ~~[its complementary sequence]~~ said locus or loci in said chromosome of interest, thereby obtaining a pattern of hybridizations between said set or ~~[clones]~~ oligo- or polynucleotides and said chromosomes, and identifying said chromosome of interest by means of said hybridization pattern obtained.

Sub  
Q  
A8  
Q8

385. (Amended) A process for identifying a plurality or all of the chromosomes in a cell of interest, the process comprising the steps of:

providing sets of ~~[clones]~~ oligo- or polynucleotides, each of said set of ~~[clones]~~ oligo- or polynucleotides being specifically ~~[complementary]~~ hybridizable to ~~[at least one sequence]~~ a locus or loci in a chromosome of said cell of interest, each of said ~~[clones]~~ oligo- or polynucleotides in said sets being labeled with a different indicator molecule and each of said ~~[clones]~~ oligo- or polynucleotides comprising at least one modified nucleotide selected from the group consisting of:

- (i) a nucleotide having the formula



wherein

PM is a phosphate moiety,

SM is a furanose moiety,

BASE is a pyrimidine, purine 7-deazapurine, and

Sig is a detectable moiety,

wherein PM is attached to the furanose moiety SM at a position selected from the group consisting of the 2', [at] the 3' [or] and the 5' position, or any combination thereof [of SM when said nucleotide is a deoxyribonucleotide and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide], BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is a purine or a 7-deazapurine, and Sig is covalently attached to BASE at a position other than the C<sup>5</sup> position when BASE is a pyrimidine, at a position other

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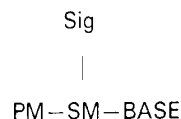
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than the C<sup>8</sup> position when BASE is a purine, and at a position other than the C<sup>2</sup> position when BASE is a 7-deazapurine;

(ii) a nucleotide having the formula



wherein

PM is a phosphate moiety,

SM is a furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is a detectable moiety,

wherein PM is attached to the furanose moiety SM at a position

[independently] selected from the group consisting of the 2', 3' and 5'

positions, or any combination thereof [of SM when said nucleotide is a

ribonucleotide, and at a position independently selected from the 3' and 5'

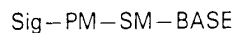
positions when said nucleotide is a deoxyribonucleotide], BASE is attached

to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or

the N<sup>9</sup> position when BASE is a purine or 7-deazapurine, and Sig is covalently

attached SM directly or through a linkage group; and

(iii) a nucleotide having the formula



wherein

PM is a phosphate moiety,

SM is a furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is detectable moiety,

wherein PM is attached to the furanose moiety SM at a position selected

from the group consisting of the 2', the 3' [or] and the 5' position, or any

combination thereof [of SM when said nucleotide is a deoxyribonucleotide

and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide],

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BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is purine, and Sig is covalently attached to PM;

fixing the chromosomes from or in said cell;

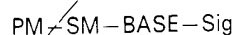
contacting said fixed chromosomes under hybridizing conditions with said sets of [clones] oligo- or polynucleotides, thereby permitting hybridization of said sets of [clones] oligo- or polynucleotides to [any of their complementary sequences] the locus or loci in said chromosomes; and

detecting any signal generated by each of said different indicator molecules in said sets of [clones] oligo- or polynucleotides which have hybridized to [their complementary sequences] the locus or loci in said chromosomes, thereby identifying [each] any one of [said] the chromosomes in said cell of interest.

390. (Amended) A process for determining the number of chromosomes in an interphase cell of interest, the process comprising the steps of:

providing sets of [clones] oligo- or polynucleotides, each of said set of [clones] oligo- or polynucleotides being specifically complementary to or specifically hybridizable with at least one [sequence] locus or loci in a chromosome of said interphase cell of interest and each of said [clones] oligo- or polynucleotides in said sets comprising at least one modified nucleotide selected from the group consisting of:

- (i) a nucleotide having the formula



wherein

PM is a phosphate moiety,

SM is a furanose moiety,

BASE is a pyrimidine, purine 7-deazapurine, and

Sig is a detectable moiety,

wherein PM is attached to the furanose moiety SM at a position selected from the group consisting of the 2', [at] the 3' [or] and the 5' position, or

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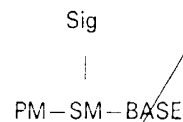
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*MXG  
cont  
Sub  
Q11*

any combination thereof [of SM when said nucleotide is a deoxyribonucleotide and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide], BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is a purine or a 7-deazapurine, and Sig is covalently attached to BASE at a position other than the C<sup>5</sup> position when BASE is a pyrimidine, at a position other than the C<sup>8</sup> position when BASE is a purine, and at a position other than the C<sup>7</sup> position when BASE is a 7-deazapurine;

(ii) a nucleotide having the formula



wherein

PM is a phosphate moiety,

SM is a furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

Sig is a detectable moiety,

wherein PM is attached to the furanose moiety SM at a position

[independently] selected from the group consisting of the 2', 3', and 5'

positions, or any combination thereof [of SM when said nucleotide is a

ribonucleotide, and at a position independently selected from the 3' and 5'

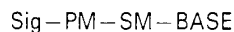
positions when said nucleotide is a deoxyribonucleotide], BASE is attached

to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or

the N<sup>9</sup> position when BASE is a purine or 7-deazapurine, and Sig is covalently

attached SM directly or through a linkage group; and

(iii) a nucleotide having the formula



wherein

PM is a phosphate moiety,

SM is a furanose moiety,

BASE is a pyrimidine, purine or 7-deazapurine, and

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Sig is detectable moiety,

wherein PM is attached to the furanose moiety SM at a position selected from the group consisting of the 2', the 3' [or] and the 5' position, or any combination thereof [of SM when said nucleotide is a deoxynucleotide and at the 2', 3' or 5' position when said nucleotide is a ribonucleotide], BASE is attached to the 1' position of SM from the N<sup>1</sup> position when BASE is a pyrimidine or the N<sup>9</sup> position when BASE is purine, and Sig is covalently attached to PM;

contacting said interphase cell under hybridizing conditions with said sets of [clones] oligo- or polynucleotides, thereby permitting hybridization of said sets of [clones] oligo- or polynucleotides to any of [their complementary sequences] the locus or loci in said chromosomes;

detecting any signals generated by each of said sets of [clones] oligo- or polynucleotides hybridized to [their complementary sequences] the locus or loci in said chromosomes, to obtain a pattern of generated signals; and comparing each generated signal with other generated signals in said pattern, thereby determining the number of chromosomes in said interphase cell of interest.

Add new claims 401-512 as follows:

-- 401. (NEW) The process of claim 376, wherein said contacting step, there are more than one oligo- or polynucleotide and each oligo- or polynucleotide is labeled with the same or a different indicator molecule. --

-- 402. (NEW) The process of claims 381 or 390, wherein said providing step each oligo- or polynucleotide is labeled with the same or a different indicator molecule. --

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-- 403. (NEW) The process of claims 381, 385 or 390, wherein said one or more oligo- or polynucleotides comprise a clone or clones or DNA fragments derived from said particular chromosome. --

-- 404. (NEW) The process of claims 329, 348 or 373, wherein said providing step each of said nucleic acid fragments is labeled with the same or a different indicator molecule. --

ML  
cont  
-- 405. (NEW) The process of claim 331, wherein said providing step each of said nucleic acid fragments is labeled with the same or a different indicator molecule. --

-- 406. (NEW) The process of claim 332, wherein said providing step each of said nucleic acid fragments is labeled with the same or a different indicator molecule. --

-- 407. (NEW) The process of claims 329, 348 or 373, wherein said fragments have been obtained or generated by a nucleic acid sequencing step or technique. --

-- 408. (NEW) The process of claims 331, wherein said fragments have been obtained or generated by a nucleic acid sequencing step or technique. --

-- 409. (NEW) The process of claims 332, wherein said fragments have been obtained or generated by a nucleic acid sequencing step or technique. --

-- 410. (NEW) The process according to claim 331, wherein Sig comprises at least three carbon atoms. --

-- 411. (NEW) The process according to claim 332, wherein A comprises at least three carbon atoms. --

-- 412. (NEW) The process according to claim 348, wherein Sig or A comprises at least three carbon atoms. --

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-- 413. (NEW) The process according to claim 331, wherein Sig comprises a monosaccharide, polysaccharide or an oligosaccharide. --

-- 414. (NEW) The process according to claim 332, wherein A comprises a monosaccharide, polysaccharide or an oligosaccharide. --

-- 415. (NEW) The process according to claim 348, wherein Sig or A comprises a monosaccharide, polysaccharide or an oligosaccharide. --

MI  
cont  
-- 416. (NEW) The process according to claim 331, wherein Sig comprises a member selected from the group consisting of biotin, iminobiotin, an electron dense component, a magnetic component, an enzyme, a hormone component, a radioactive component, a metal-containing component, a fluorescent component, a chemiluminescent component, an antigen, a hapten, an antibody component and a chelating component. --

-- 417. (NEW) The process according to claim 332, wherein A comprises a member selected from the group consisting of biotin, iminobiotin, an electron dense component, a magnetic component, an enzyme, a hormone component, a radioactive component, a metal-containing component, a fluorescent component, a chemiluminescent component, an antigen, a hapten, an antibody component and a chelating component. --

-- 418. (NEW) The process according to claim 348, wherein Sig or A comprises a member selected from the group consisting of biotin, iminobiotin, an electron dense component, a magnetic component, an enzyme, a hormone component, a radioactive component, a metal-containing component, a fluorescent component, a chemiluminescent component, an antigen, a hapten, an antibody component and a chelating component. --

-- 419. (NEW) The process according to claim 416, wherein Sig comprises an electron dense component. --



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-- 420. (NEW) The process according to claim 417, wherein A comprises an electron dense component. --

-- 421. (NEW) The process according to claim 418, wherein Sig or A comprises an electron dense component. --

-- 422. (NEW) The process according to claim 419, wherein said electron dense component comprises ferritin. --

-- 423. (NEW) The process according to claim 420, wherein said electron dense component comprises ferritin. --

-- 424. (NEW) The process according to claim 421, wherein said electron dense component comprises ferritin. --

-- 425. (NEW) The process according to claim 416, wherein Sig comprises a magnetic component. --

-- 426. (NEW) The process according to claim 417, wherein A comprises a magnetic component. --

-- 427. (NEW) The process according to claim 418, wherein Sig or A comprises a magnetic component. --

-- 428. (NEW) The process according to claim 425, wherein said magnetic component comprises magnetic oxide or magnetic iron oxide. --

-- 429. (NEW) The process according to claim 426, wherein said magnetic component comprises magnetic oxide or magnetic iron oxide. --

-- 430. (NEW) The process according to claim 427, wherein said magnetic component comprises magnetic oxide or magnetic iron oxide. --

M11  
cont

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-- 431. (NEW) The process according to claim 428, wherein said magnetic component comprises magnetic beads. --

-- 432. (NEW) The process according to claim 429, wherein said magnetic component comprises magnetic beads. --

-- 433. (NEW) The process according to claim 430, wherein said magnetic component comprises magnetic beads. --

MI  
cont  
-- 434. (NEW) The process according to claim 331, wherein Sig comprises a sugar residue and the sugar residue is complexed with or attached to a sugar binding protein or a polysaccharide binding protein. --

-- 435. (NEW) The process according to claim 332, wherein A comprises a sugar residue and the sugar residue is complexed with or attached to a sugar binding protein or a polysaccharide binding protein. --

-- 436. (NEW) The process according to claim 348, wherein Sig or A comprises a sugar residue and the sugar residue is complexed with or attached to a sugar binding protein or a polysaccharide binding protein. --

-- 437. (NEW) The process according to claim 434, wherein the binding protein comprises a lectin. --

-- 438. (NEW) The process according to claim 435, wherein the binding protein comprises a lectin. --

-- 439. (NEW) The process according to claim 436, wherein the binding protein comprises a lectin. --

03  
-- 440. (NEW) The process according to claim 437, wherein the lectin comprises Concanavalin A. --

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sub  
Q3  
-- 441. (NEW) The process according to claim 438, wherein the lectin comprises Concanavalin A. --

-- 442. (NEW) The process according to claim 439, wherein the lectin comprises Concanavalin A. --

-- 443. (NEW) The process according to claim 437, wherein the lectin is conjugated to ferritin. --

M11  
cont  
-- 444. (NEW) The process according to claim 438, wherein the lectin is conjugated to ferritin. --

-- 445. (NEW) The process according to claim 439, wherein the lectin is conjugated to ferritin. --

-- 446. (NEW) The process according to claim 416, wherein Sig comprises an enzyme. --

-- 447. (NEW) The process according to claim 417, wherein A comprises an enzyme. --

-- 448. (NEW) The process according to claim 418, wherein Sig or A comprises an enzyme. --

-- 449. (NEW) The process according to claim 446, wherein the enzyme is selected from the group consisting of alkaline phosphatase, acid phosphatase,  $\beta$ -galactosidase, ribonuclease, glucose oxidase and peroxidase, or a combination thereof. --

-- 450. (NEW) The process according to claim 447, wherein the enzyme is selected from the group consisting of alkaline phosphatase, acid phosphatase,  $\beta$ -galactosidase, ribonuclease, glucose oxidase and peroxidase, or a combination thereof. --

Enz-5(D8)(C2)

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-- 451. (NEW) The process according to claim 448, wherein the enzyme is selected from the group consisting of alkaline phosphatase, acid phosphatase,  $\beta$ -galactosidase, ribonuclease, glucose oxidase and peroxidase, or a combination thereof. --

-- 452. (NEW) The process according to claim 416, wherein Sig comprises a hormone. --

-- 453. (NEW) The process according to claim 417, wherein A comprises a hormone. --

-- 454. (NEW) The process according to claim 418, wherein Sig or A comprises a hormone. --

-- 455. (NEW) The process according to claim 416, wherein Sig comprises a radioactive isotope. --

-- 456. (NEW) The process according to claim 417, wherein A comprises a radioactive isotope. --

-- 457. (NEW) The process according to claim 418, wherein Sig or A comprises a radioactive isotope. --

-- 458. (NEW) The process according to claim 416, wherein Sig comprises a metal-containing component. --

-- 459. (NEW) The process according to claim 417, wherein A comprises a metal-containing component. --

-- 460. (NEW) The process according to claim 418, wherein Sig or A comprises a metal-containing component. --

-- 461. (NEW) The process according to claim 458, wherein said metal-containing component is catalytic. --

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-- 462. (NEW) The process according to claim 459, wherein said metal-containing component is catalytic. --

-- 463. (NEW) The process according to claim 460, wherein said metal-containing component is catalytic. --

MH  
cont  
-- 464. (NEW) The process according to claim 416, wherein Sig comprises a fluorescent component. --

-- 465. (NEW) The process according to claim 417, wherein A comprises a fluorescent component. --

-- 466. (NEW) The process according to claim 418, wherein Sig or A comprises a fluorescent component. --

-- 467. (NEW) The process according to claim 464, wherein the fluorescent component is selected from the group consisting of fluorescein, rhodamine and dansyl. --

-- 468. (NEW) The process according to claim 465, wherein the fluorescent component is selected from the group consisting of fluorescein, rhodamine and dansyl. --

-- 469. (NEW) The process according to claim 466, wherein the fluorescent component is selected from the group consisting of fluorescein, rhodamine and dansyl. --

-- 470. (NEW) The process according to claim 416, wherein Sig comprises a chemiluminescent component. --

-- 471. (NEW) The process according to claim 417, wherein A comprises a chemiluminescent component. --

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-- 472. (NEW) The process according to claim 418, wherein Sig or A comprises a chemiluminescent component. --

-- 473. (NEW) The process according to claim 331 wherein Sig comprises an antigenic or hapten component capable of complexing with an antibody specific to the component. --

-- 474. (NEW) The process according to claim 332, wherein A comprises an antigenic or hapten component capable of complexing with an antibody specific to the component. --

-- 475. (NEW) The process according to claim 348, wherein Sig or A comprises an antigenic or hapten component capable of complexing with an antibody specific to the component. --

-- 476. (NEW) The process according to claim 416, wherein Sig comprises an antibody component. --

-- 477. (NEW) The process according to claim 417, wherein A comprises an antibody component. --

-- 478. (NEW) The process according to claim 418, wherein Sig or A comprises an antibody component. --

-- 479. (NEW) The process according to claim 416, wherein Sig comprises a chelating component. --

-- 480. (NEW) The process according to claim 417, wherein Sig comprises a chelating component. --

-- 481. (NEW) The process according to claim 418, wherein Sig or A comprises a chelating component. --

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-- 482. (NEW) The process according to claim 331, wherein Sig is detectable when said modified nucleotides are contained in a double-stranded ribonucleic or deoxyribonucleic acid duplex. --

-- 483. (NEW) The process according to claim 332, wherein A is detectable when said modified nucleotides are contained in a double-stranded ribonucleic or deoxyribonucleic acid duplex. --

MI  
cont  
-- 484. (NEW) The process according to claim 348, wherein Sig or A is detectable when said modified nucleotides are contained in a double-stranded ribonucleic or deoxyribonucleic acid duplex. --

-- 485. (NEW) The process according to claim 331, wherein Sig is detectable when it is attached to the nucleotide directly or through a linkage group. --

-- 486. (NEW) The process according to claims 332, wherein A is detectable when it is attached to the nucleotide directly or through a linkage group. --

-- 487. (NEW) The process according to claims 348, wherein Sig or A is detectable when it is attached to the nucleotide directly or through a linkage group. --

-- 488. (NEW) The process according to claim 485, wherein said linkage group does not interfere substantially with the characteristic ability of Sig to form a detectable signal. --

-- 489. (NEW) The process according to claim 486, wherein said linkage group does not interfere substantially with the characteristic ability of A to form a detectable signal. --

-- 490. (NEW) The process according to claim 487, wherein said linkage group does not interfere substantially with the characteristic ability of Sig or A to form a detectable signal. --

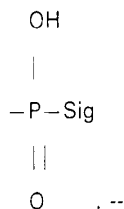
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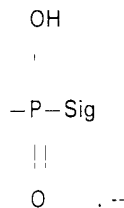
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-- 491. (NEW) The process according to claim 331, wherein Sig in said nucleotide (iii) is covalently attached to PM via the chemical linkage

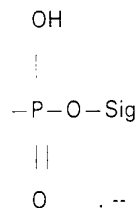


M11  
cont

-- 492. (NEW) The process according to claims 337 or 348, wherein Sig in said nucleotide (iii) is covalently attached to PM via the chemical linkage



-- 493. (NEW) The process according to claim 331, wherein Sig in said nucleotide (iii) is covalently attached to PM via the chemical linkage





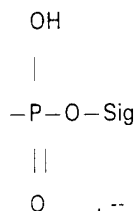
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-- 494. (NEW) The process according to claims 337 or 348, wherein Sig in said nucleotide (iii) is covalently attached to PM via the chemical linkage



-- 495. (NEW) The process according to claim 331, wherein said nucleic acid fragments are terminally ligated or attached to a polypeptide. --

-- 496. (NEW) The process according to claims 332, wherein said nucleic acid fragments are terminally ligated or attached to a polypeptide. --

-- 497. (NEW) The process according to claim 348, wherein said nucleic acid fragments are terminally ligated or attached to a polypeptide. --

-- 498. (NEW) The process according to claims 495, wherein the polypeptide comprises a polylysine. --

-- 499. (NEW) The process according to claims 496, wherein the polypeptide comprises a polylysine. --

-- 500. (NEW) The process according to claims 497, wherein the polypeptide comprises a polylysine. --

-- 501. (NEW) The process according to claims 495, wherein the polypeptide comprises at least one member selected from the group consisting of avidin, streptavidin or anti-Sig immunoglobulin. --

-- 502. (NEW) The process according to claims 496, wherein the polypeptide comprises at least one member selected from the group consisting of avidin, streptavidin or anti-Sig immunoglobulin. --

Enz-5(D8)(C2)

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-- 503. (NEW) The process according to claims 497, wherein the polypeptide comprises at least one member selected from the group consisting of avidin, streptavidin or anti-Sig immunoglobulin. --

ML  
cont  
-- 504. (NEW) The process according to claim 331, wherein Sig comprises a ligand and the polypeptide comprises an antibody thereto. --

-- 505. (NEW) The process according to claim 332, wherein A comprises a ligand and the polypeptide comprises an antibody thereto. --

-- 506. (NEW) The process according to claim 348, wherein Sig or A comprises a ligand and the polypeptide comprises an antibody thereto. --

-- 507. (NEW) The process according to claim 495, further comprising a moiety which can be detected when a complex is formed between Sig and said polypeptide, said moiety being selected from the group consisting of biotin, iminobiotin, an electron dense component, a magnetic component, an enzyme, a hormone component, a radioactive component, a metal-containing component, a fluorescent component, a chemiluminescent component, an antigen, a hapten, an antibody component and a chelating component. --

-- 508. (NEW) The process according to claim 496, further comprising a moiety which can be detected when a complex is formed between A and said polypeptide, said moiety being selected from the group consisting of biotin, iminobiotin, an electron dense component, a magnetic component, an enzyme, a hormone component, a radioactive component, a metal-containing component, a fluorescent component, a chemiluminescent component, an antigen, a hapten, an antibody component and a chelating component. --

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Amendment Under 37 C.F.R. §1.116 - May 1, 1999)

-- 509. (NEW) The process according to claim 497, further comprising a moiety which can be detected when a complex is formed between Sig and said polypeptide, said moiety being selected from the group consisting of biotin, iminobiotin, an electron dense component, a magnetic component, an enzyme, a hormone component, a radioactive component, a metal-containing component, a fluorescent component, a chemiluminescent component, an antigen, a hapten, an antibody component and a chelating component. --

-- 510. (NEW) The process of claim 284, further comprising one or more washing steps. --

-- 511. (NEW) The process of claim 376, wherein said contacting step the detectable moiety Sig in said modified nucleotide comprises a member selected from the group consisting of biotin, iminobiotin, an electron dense component, a magnetic component, an enzyme, a hormone component, a radioactive component, a metal-containing component, a fluorescent component, a chemiluminescent component, an antigen, a hapten, an antibody component and a chelating component. --

-- 512. (NEW) The process of claims 381, 385 or 390, wherein said providing step the detectable moiety Sig in said modified nucleotide comprises a member selected from the group consisting of biotin, iminobiotin, an electron dense component, a magnetic component, an enzyme, a hormone component, a radioactive component, a metal-containing component, a fluorescent component, a chemiluminescent component, an antigen, a hapten, an antibody component and a chelating component. --

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